

## Evaluation of VITEK 2 Rapid Identification and Susceptibility Testing System against Gram-Negative Clinical Isolates

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A total of 281 strains of miscellaneous members of the family *Enterobacteriaceae*, *Pseudomonas aeruginosa*, and other gram-negative bacteria were evaluated by use of identification tests with the VITEK 2 system (bioMérieux) and an API identification system (bioMérieux). A total of 237 (95%) strains were correctly identified to the species level. Only six (2.1%) strains were misidentified, and eight (2.8%) strains were not identified. Among 14 strains with discrepant identifications, 8 (57.1%) strains were nonfermenters. The susceptibilities of 228 strains to 11 antibiotics including amikacin, netilmicin, tobramycin, gentamicin, ciprofloxacin, imipenem, meropenem, ceftazidime, cefepime, piperacillin, and piperacillin in combination with tazobactam were tested with the VITEK 2 AST-No. 12 card and by the broth microdilution (MB) method, according to NCCLS guidelines, as a reference. For the 2,508 organism-antibiotic combinations, the rates at which duplicate MICs correlated within  $\pm 1$  dilution ranged from 84.2 to 95.6%. Only 13 (0.5%) and 10 (0.4%) of the susceptibility tests gave major errors (resistant with the VITEK 2 system but sensitive by the MB method) and very major errors (sensitive with the VITEK 2 system but resistant by the MB method), respectively. Both VITEK 2 ID-GNB (an identification system) and VITEK 2 AST-No. 12 (a susceptibility testing system) card systems gave rapid, reliable, and highly reproducible results.

Automated bacterial identification and susceptibility testing systems have been developed and commercialized for more than two decades, but only a few of them are available on the market (1, 2). The VITEK 2 system (bioMérieux), which uses a new fluorescence-based technology, was evaluated for the identification and susceptibility testing of gram-negative clinical isolates.

Clinical isolates were collected from 1996 to 1999 in the Prince of Wales Hospital, Hong Kong, People's Republic of China, and were stored in cryotubes at  $-70^{\circ}\text{C}$ . The stock culture strains were then subcultured onto MacConkey agar plates to check their purity. The turbidity of the bacterial suspensions was adjusted with a densitometer to match that of a McFarland 0.5 standard in 0.45% sterile sodium chloride solution. The time interval between suspension preparation and card filling was less than 30 min to avoid changes in turbidity. Afterward, the VITEK 2 ID-GNB cards, AST-No. 12 cards, and bacterial suspension were manually loaded into the VITEK 2 system. Each test card was automatically filled with a bacterial suspension, sealed, and incubated for 3 h. During this period, the cards were read by kinetic fluorescence measurement every 15 min. The VITEK 2 system software first analyzed the data and then reported the results automatically.

A total of 281 strains of miscellaneous members of the family *Enterobacteriaceae* ( $n = 173$ ), *Pseudomonas aeruginosa* ( $n = 23$ ), and other gram-negative bacteria ( $n = 85$ ) were tested with the VITEK 2 ID-GNB cards. The reference identification was obtained with the API 20E system (bioMérieux) (3).

The susceptibilities of 228 strains to 11 antibiotics were tested with the VITEK 2 AST-No. 12 card and by the broth

microdilution (MB) method, according to NCCLS guidelines, as a reference (4). The reference MIC was determined by the MB method (MIC-2000 System; Dynatech, McLean, Va.) with Muller-Hinton broth (Oxoid, Basingstoke, United Kingdom)

TABLE 1. Organisms with correct identification with the VITEK 2 system

| Organism name   | No. of strains |
|---|----------------|
| <i>Acinetobacter baumannii</i> .....                        | 31             |
| <i>Acinetobacter junii</i> .....                            | 1              |
| <i>Acinetobacter lwoffii</i> .....                          | 1              |
| <i>Aeromonas caviae</i> .....                               | 1              |
| <i>Aeromonas hydrophila</i> .....                           | 8              |
| <i>Aeromonas sobria</i> .....                               | 3              |
| <i>Alcaligenes faecalis</i> .....                           | 1              |
| <i>Alcaligenes xylosoxidans</i> .....                       | 5              |
| <i>Burkholderia cepacia</i> .....                           | 2              |
| <i>Chryseobacterium indologenes</i> .....                   | 1              |
| <i>Chryseobacterium meningosepticum</i> .....               | 3              |
| <i>Citrobacter freundii</i> .....                           | 1              |
| <i>Citrobacter koseri</i> .....                             | 3              |
| <i>Edwardsiella tarda</i> .....                             | 1              |
| <i>Enterobacter cloacae</i> .....                           | 13             |
| <i>Escherichia coli</i> .....                               | 62             |
| <i>Klebsiella oxytoca</i> .....                             | 8              |
| <i>Klebsiella pneumoniae</i> subsp. <i>pneumoniae</i> ..... | 29             |
| <i>Morganella morganii</i> .....                            | 2              |
| <i>Plesiomonas shigelloides</i> .....                       | 1              |
| <i>Proteus mirabilis</i> .....                              | 10             |
| <i>Proteus vulgaris</i> .....                               | 6              |
| <i>Providencia rettgeri</i> .....                           | 4              |
| <i>Providencia stuartii</i> .....                           | 2              |
| <i>Pseudomonas aeruginosa</i> .....                         | 23             |
| <i>Pseudomonas luteola</i> .....                            | 1              |
| <i>Ralstonia pickettii</i> .....                            | 1              |
| <i>Salmonella</i> spp. ....                                 | 12             |
| <i>Salmonella typhi</i> .....                               | 3              |
| <i>Serratia marcescens</i> .....                            | 13             |
| <i>Stenotrophomonas maltophilia</i> .....                   | 15             |
| Total .....   | 267            |

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TABLE 2. Organisms with discrepant identifications with the VITEK 2 system

| Identity with API 20E system   | Identity with VITEK 2 system        | VITEK 2 system confidence level |
|--------------------------------|-------------------------------------|---------------------------------|
| <i>Acinetobacter baumannii</i> | Unidentified organism               | Unidentified organism           |
| <i>Acinetobacter lwoffii</i>   | <i>Stenotrophomonas maltophilia</i> | Excellent identification        |
| <i>Acinetobacter junii</i>     | Unidentified organism               | Unidentified organism           |
| <i>Aeromonas caviae</i>        | Unidentified organism               | Unidentified organism           |
| <i>Alcaligenes faecalis</i>    | Unidentified organism               | Unidentified organism           |
| <i>Citrobacter koseri</i>      | Unidentified organism               | Unidentified organism           |
| <i>Escherichia coli</i>        | Unidentified organism               | Unidentified organism           |
| <i>Klebsiella oxytoca</i>      | <i>Citrobacter koseri</i>           | Acceptable identification       |
| <i>Pseudomonas fluorescens</i> | <i>Pseudomonas aeruginosa</i>       | Low discrimination              |
| <i>Pseudomonas putida</i>      | Unidentified organism               | Unidentified organism           |
| <i>Pseudomonas stutzeri</i>    | <i>Ralstonia pickettii</i>          | Very good identification        |
| <i>Pseudomonas stutzeri</i>    | <i>Pseudomonas aeruginosa</i>       | Low discrimination              |
| <i>Serratia liquefaciens</i>   | <i>Enterobacter cloacae</i>         | Good identification             |
| <i>Vibrio vulnificus</i>       | Unidentified organism               | Unidentified organism           |

with an inoculum size of  $10^5$  CFU/ml. The following antibiotics were supplied as powders of stated potency and were obtained from the indicated companies: amikacin, Bristol-Myers Squibb; netilmicin, Schering Plough; tobramycin, Sigma; gentamicin, Sigma; ciprofloxacin, Bayer; imipenem, Merck Sharp & Dohme; meropenem, AstraZeneca; ceftazidime, Glaxo Wellcome; cefepime, Bristol-Myers Squibb; piperacillin, Wyeth; and piperacillin in combination with tazobactam, Wyeth. *Escherichia coli* ATCC 25922 and *Pseudomonas aeruginosa* ATCC 27853 were incorporated as quality control strains. All strains were allowed to be tested only if the results were correct for all quality control strains. All strains with discrepant results were retested in duplicate by both methods.

Among 281 strains (85 nonfermenters) tested, 267 (95%) strains were correctly identified to the species level (Table 1). Only six (2.1%) strains were misidentified, and eight (2.8%) strains were not identified. Among the strains with discrepant results, eight were nonfermenters (Table 2). The VITEK 2 AST-No. 12 card gave highly reproducible antibiotic susceptibility testing results. For the 2,508 organism-antibiotic combinations (composed of 228 strains tested against 11 antibiotics), the rates at which duplicate MICs correlated within  $\pm 1$  dilution ranged from 84.2 to 95.6%. Only 13 (0.5%) and 10 (0.4%) tests gave major errors (resistant with the VITEK 2 system but sensitive by the MB method) and very major errors (sensitive with the VITEK 2 system but resistant by the MB method), respectively (Table 3).

In the present study, 8 of 14 (57.1%) organisms with discrepant identifications with the VITEK 2 system (either misidentified or not identified) were nonenteric bacteria such as *Pseudomonas* spp. (4), *Acinetobacter* spp. (3), and *Alcaligenes* (1). The slower rate of metabolism of nonenteric bacteria could cause weaker fluorescent biochemical reactions in the reaction wells of VITEK 2 ID-GNB cards, and this may cause more discrepant identifications with the VITEK 2 system. This evaluation showed that VITEK 2 system could identify members of the family *Enterobacteriaceae* and fermenters better. An evaluation performed by Funke et al. (1) also showed that the rate of rapid and reliable identification of gram-negative isolates to the species level with the VITEK 2 system is 84.7%. However, both studies revealed that improvements should be

made to identify nonenteric organisms and nonfermenters with slower metabolisms.

After comparison of the susceptibility testing results obtained with the VITEK 2 AST-No. 12 cards with those obtained by the MB method, major errors (resistant with the VITEK 2 system but sensitive by the MB method) and very major errors (sensitive with the VITEK 2 system but resistant by the MB method) were randomly distributed among the tests with the 11 drugs listed. No specific pattern was found. Another study performed by Traczewski et al. (M. M. Traczewski, A. L. Barry, S. D. Brown, J. A. Hingler, D. A. Bruckner, and D. F. Sahn, Abstr. 98th Gen. Meet. Am Soc. Microbiol. 1998, p. 27–28, 1998) showed that the rate of agreement of MICs ( $\pm 1$  dilution) from the susceptibility testing results for all cephalosporins tested against gram-negative isolates was over 90%, which is similar to those obtained in our study, which ranged between 84.7 and 95.6%.

During the evaluation, the overall performance of the VITEK 2 system was satisfactory except for a few minor mechanical defects. Several advantages of the VITEK 2 system can be mentioned. First, it is a closed system that can avoid unwanted cross-contamination or environmental contamination. Second, it has a reliable recheck system that can detect and immediately cease operation of the VITEK 2 system if a specimen card is misplaced on the specimen cartridge. Third, the VITEK 2 system is able to handle dozens of specimens automatically at the same time. It is also easy for laboratory staff to prepare and load bacterial specimens. The decreased turnaround and hand-on times greatly improve the efficiencies of routine clinical laboratories. In conclusion, both the VITEK 2 ID-GNB (an identification system) and VITEK 2 AST-No. 12 (a susceptibility testing system) card systems gave rapid, reliable, and highly reproducible results.

TABLE 3. Agreement of MB method and VITEK system for 228 clinical isolates

| Antibiotic              | No. (%) of strains with:   |                                 |                    |                    |                         |
|-------------------------|----------------------------|---------------------------------|--------------------|--------------------|-------------------------|
|                         | MIC agreement <sup>a</sup> | Category agreement <sup>b</sup> | Error              |                    |                         |
|                         |                            |                                 | Minor <sup>c</sup> | Major <sup>d</sup> | Very major <sup>e</sup> |
| Amikacin                | 215 (94.3)                 | 2                               | 10                 | 0                  | 1                       |
| Gentamicin              | 218 (95.6)                 | 5                               | 3                  | 1                  | 1                       |
| Tobramycin              | 213 (93.4)                 | 5                               | 9                  | 1                  | 0                       |
| Netilmicin              | 211 (92.5)                 | 9                               | 5                  | 3                  | 0                       |
| Ciprofloxacin           | 209 (91.7)                 | 17                              | 2                  | 0                  | 0                       |
| Imipenem                | 192 (84.2)                 | 29                              | 7                  | 0                  | 0                       |
| Meropenem               | 212 (93.0)                 | 14                              | 1                  | 0                  | 1                       |
| Ceftazidime             | 211 (92.5)                 | 7                               | 5                  | 4                  | 1                       |
| Cefepime                | 210 (92.1)                 | 11                              | 5                  | 1                  | 1                       |
| Piperacillin            | 204 (89.5)                 | 17                              | 3                  | 2                  | 2                       |
| Piperacillin-tazobactam | 193 (84.7)                 | 29                              | 2                  | 1                  | 3                       |

<sup>a</sup> MIC agreement ( $\pm 1$  dilution).

<sup>b</sup> Interpretive category agreement, although MICs differed by  $>1$  dilution.

<sup>c</sup> Minor errors (susceptible or resistant with the VITEK 2 system and intermediate by the reference test or intermediate with the VITEK 2 system and susceptible or resistant by the reference test).

<sup>d</sup> Major errors (resistant with the VITEK 2 system and susceptible by the reference test).

<sup>e</sup> Very major errors (susceptible with the VITEK 2 system and resistant by the reference test).

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